K002024

NOV 22 2000

Innovative Diagnostic Solutions

PanBio InDx, Inc.
1756 Sulphur Spring Road
Baltimore, Maryland 21227
USA

Tel: 410-737-8500
Fax: 410-536-1212
Toll Free: 1-800-832-4639
Internet: www.indxdi.com
E-mail: indx2@erols.com

510(k) SUMMARY
SUMMARY OF SAFETY AND EFFECTIVENESS DATA

NAME AND LOCATION OF MANUFACTURER

PAN BIO-INDX, INC.
1756 Sulphur Spring Road
Baltimore, Maryland 21227

NAME OF CONTACT PERSON

Helene Paxton, M.S., M.T.
Vice President for Regulatory Affairs, Manufacturing, Research and Development
PAN BIO-INDX, INC.

DATE OF PREPARATION OF SUMMARY

November 16, 2000

TRADE NAME OF THE DEVICE

Pan Bio InDx^{R} Dip-S-Ticks^{R} Leptospirosis Test for the Detection of IgM Antibodies To Leptospira biflexa

COMMON NAME OF THE DEVICE

Pan Bio InDx IgM Dip-S-Ticks Leptospirosis Test

CLASSIFICATION NAME

CFR 866.3350, Leptospira spp. Serological reagents, Product Code: GRY

LEGALLY MARKETED (PREDICATE) DEVICE TO WHICH THE MANUFACTURER IS CLAIMING SUBSTANTIAL EQUIVALENCE

Indirect Hemagglutination (IHA) Test for the Detection of Human Antibodies to the Genus Leptospira, Manufactured by MRL Diagnostics, Cypress, California 90630

DESCRIPTION OF THE DEVICE

The PanBio InDx IgM Dip-S-Ticks Leptospirosis Test utilizes an enzyme-linked immunoassay (EIA) dot technique for the detection of IgM antibodies. Nitrocellulose strips are provided with *Leptospira biflexa*, serovar Patoc 1 strain antigens dispensed as discrete dots. *Leptospira* organisms are grown in EMJH medium to a final concentration of 10^{9}/ml. The culture is inactivated by formalin, treated in a boiling water bath for 30 minutes and clarified by centrifugation. Candidate antigen preparations are subsequently assayed for potency and the absence of non-specific binding, by comparisons with well-characterized positive and negative clinical specimens, prior to application to the nitrocellulose strips.

The test is performed by first adding the test specimen to a reaction cuvette. The antigen-containing assay strip is inserted, allowing patient antibodies reactive with the test antigens to bind to the strip's solid support membrane.

Alkaline phosphatase-conjugated goat anti-human IgM antibodies are allowed to react with bound patient antibodies. Finally, the strip is transferred to an enzyme substrate reagent, which reacts with bound alkaline phosphatase to produce an easily seen blue to purple-colored, distinct spot.

INTENDED USE OF THE DEVICE

The PanBio InDx® IgM Dip-S-Ticks® Leptospirosis test is a qualitative enzyme immunoassay that specifically detects IgM antibodies to *Leptospira biflexa* (serovar *patoc 1*). This test is presumptive for the laboratory diagnosis of leptospirosis. The IgM Dip-S-Ticks Test may be used to evaluate a single specimen or paired specimens to detect sero-conversion. The test is intended for use in serum, plasma, heparinized whole blood or finger stick capillary blood.

SUMMARY OF TECHNICAL CHARACTERISTICS OF THE DEVICE COMPARED TO THE PREDICATE DEVICE

The manufacturer's device is intended for the detection of the IgM leptospirosis antibodies using an ELISA format. Antigen is imbedded in a nitrocellulose matrix in a series of 4-fold dilutions. The device is intended for use with multiple matrices, including serum, plasma, whole blood and finger-stick capillary blood. The predicate device consists of human type "O" erythrocytes coated with genus-specific leptospiral antigens. The presence of antibody in the patient's serum causes agglutination of red cells. The test may detect IgM and IgG leptospirosis antibody and is intended for use with serum samples only. Both tests utilize *Leptospira biflexa* (serovar *patoc 1*) antigens.

NON-CLINICAL TESTS SUPPORTING A DETERMINATION OF SUBSTANTIAL EQUIVALENCE

Expected Value Data:

Two independent studies, one in a rural and the other in an urban area of Maryland, were conducted with samples from asymptomatic normal donors. In the first study, sera were collected from 216 donors during the period of May through October of 1997. These rural samples were from the Maryland Eastern Shore with no known prevalence for leptospirosis. Sera from 207 of these donors were analyzed with the IgM Dip-S-Ticks Leptospirosis Test. Two sera (1.0%) were positive and 205 (99.0%) were negative. In the second study of 100 urban samples from normal donors, 97 were negative with the IgM Dip-S-Ticks Leptospirosis test and 3 were positive. However, using other leptospirosis assay methods, the frequency of positive specimens among normal donors was reported to be 0.1% in Hawaii and less than 0.001% in the continental United States.

Specificity (Cross-reactivity) Data:

This study consisted of a panel of 170 specimens from patients with diseases other than leptospirosis. Samples were selected to represent infectious and non-infectious diseases having a febrile phase that may clinically mimic or be confused with leptospirosis. The numbers of each specimen type tested are indicated in parenthesis ( ).

These were parasitic and bacterial diseases (42) including (syphilis (11) scrub typhus (10) dengue (9) mycoplasma (8), tularensis (2) toxoplasma (2). Also included were ANA positive autoimmune (35), rickettsia (9), ehrlichia (3), Lyme disease (5), Salmonella typhi (10), Chagas (4) and miscellaneous (5). Viral diseases (57) included CMV (11), EBV (12), Hepatitis (7), HIV (2), HTLV (1), HSV (10) Rubeola and Rubella (3) and VZV (11). Results of the study indicated that 163 specimens (95.9%) were negative and 7 (4.1%) were false positive when analyzed with the IgM Dip-S-Ticks test. Among false positive specimens were 1 each of ANA, HSV, syphilis, scrub typhus, dengue and 2 of Salmonella typhi. All were borderline positive, including 5 of 2.0 dots, and 2 of 2.5 dots.

Reproducibility Data:

A study was conducted to assess the inter-laboratory reproducibility of the IgM Dip-S-Ticks Leptospirosis Test. A panel of 30 coded and blinded leptospirosis positive and negative sera were analyzed in each of 3 laboratories, including 2 clinical laboratories and the manufacturer's location. Positive sera were selected in a manner to represent different time periods of leptospirosis IgM antibody reactivity. Ten sera analyzed in each laboratory were obtained from patients less than 10 days post-onset of infection and 10 of the sera were obtained 10-30 days post-onset. In addition, 10 of the sera were from negative controls.

To enable the demonstration of reproducibility throughout the entire measurement range of the IgM Dip-S-Ticks Leptospirosis Test, the 15 leptospirosis positive sera included samples in the reactivity range of 2.0-3.0 dots and the reactivity range of 3.0-4.0 dots. The 10 negative sera were in the range of less than 2.0 dots, or less than the cutoff point for a positive test. Table 1 describes the reproducibility study of positive samples in 3 independent laboratories. Negative samples were consistently non-reactive in all laboratories.

TABLE 1
LEPTOSPIROSIS POSITIVE SAMPLES

Sample Number	Days Post Onset	Reproducibility Data Obtained (Number of positive dots) Site #1	Site #2	Site #3	Mean	1 S.D.
37094	# 10	2.5	2.5	2.5	2.50	0.00
37195	# 10	4.0	4.0	4.0	4.00	0.00
37357	# 10	4.0	3.5	3.5	3.83	0.29
37414	# 10	4.0	3.5	3.0	3.50	0.50
36404	# 10	3.0	2.5	3.0	2.83	0.29
36049	# 10	2.5	2.5	2.5	2.50	0.00
36308	# 10	4.0	4.0	4.0	4.00	0.00
37103	# 10	4.0	3.5	3.5	3.67	0.29
37655	# 10	4.0	3.5	3.0	3.50	0.50
29341	# 10	3.0	2.5	3.0	2.83	0.29
36243	10-30	4.0	4.0	3.5	3.83	0.29
36291	10-30	4.0	4.0	4.0	4.00	0.00
38476	10-30	4.0	4.0	4.0	4.00	0.00
36624	10-30	4.0	4.0	4.0	4.00	0.00
37064	10-30	3.5	4.0	3.5	3.66	0.29
37085	10-30	4.0	4.0	4.0	4.00	0.00
37195	10-30	4.0	4.0	4.0	4.00	0.00
37374	10-30	4.0	4.0	4.0	4.00	0.00
37440	10-30	4.0	3.5	3.0	3.50	0.50
37521	10-30	4.0	4.0	3.5	3.83	0.29
Mean		3.7	3.6	3.5		
1 S.D.		0.53	0.59	0.53		

In a second study of reproducibility, ten masked clinical specimens were each analyzed in triplicate (n=30) for 3 test days at each of three clinical sites. The mean number of dots, standard deviation (S.D.) and coefficient of variation (CV) were determined for each specimen in each site on each test day, as shown in Table 2.

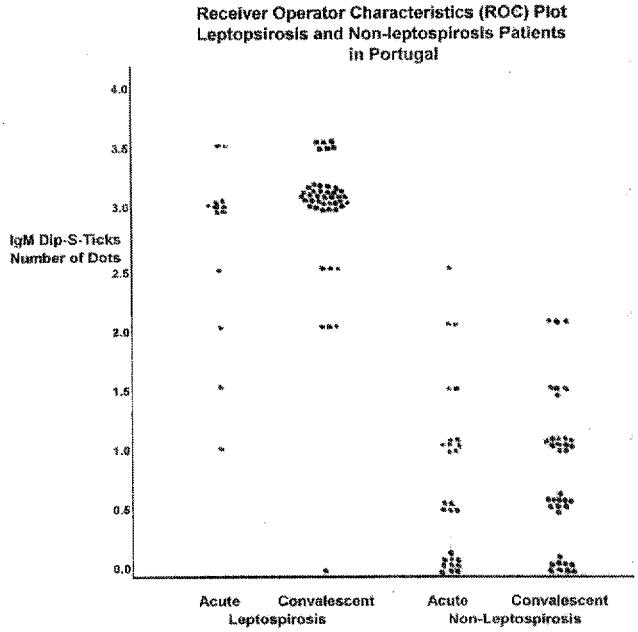
Table 2
REPRODUCIBILITY OF IgM Dip-S-Ticks TEST

	Site #1			Site #2			Site #3			
Test Days	Mean # dots	S.D.	CV (%)	Mean # dots	S.D.	CV (%)	Mean # dots	S.D.	CV (%)	N
1	2.4	0.20	8.48	2.4	0.17	7.10	2.5	0.28	11.05	30
2	2.4	0.24	10.09	2.4	0.23	9.92	2.5	0.25	9.74	30
3	2.5	0.21	8.33	2.5	0.18	7.21	2.7	0.35	13.25	30
Total	2.4	0.22	8.97	2.4	0.20	8.08	2.6	0.29	11.34	90

Assay Cutoff Analysis

The Receiver Operating Characteristic (ROC) dot-plot analysis (7) was used to examine the IgM Dip-S-Ticks test distribution of reactivity and cutoff points for acute and convalescent-phase leptospirosis and non-leptospirosis specimens analyzed in Study No. 3. The ROC plot is shown in Figure 1.

FIGURE 1



Acute-phase specimens were those obtained <10 days post-onset. Convalescent-phase specimens were those obtained 10-30 days and >30 days post-onset. Among 58 leptospirosis specimens, 13 were acute-phase and 45 were convalescent-phase. The percent positive agreement with the MAT test was 84.6% among acute-phase specimens and 97.8% among convalescent-phase specimens. Among 64 non-leptospirosis specimens, 27 were acute-phase and 37 were convalescent-phase. The percent negative agreement with the MAT test was 88.9% among acute-phase specimens and 91.9% among convalescent-phase specimens.

Since relatively few patients showed IgM Dip-S-Ticks values at the 2.0 dot cutoff point in the above study, the performance of the test at the cutoff point for all clinical study sites was determined. In this analysis, only 7.5% (55/731) specimens from symptomatic patients showed a 2.0 dot cutoff value with the IgM Dip-S-Ticks test. Twenty two of these (40%) were MAT positive and 9 (16.4%) were IHA positive at the cutoff. Therefore, the 2.0 dot reactive acute-phase specimens could be interpreted as false positives and the analysis of a second specimen collected 2-3 weeks later or use of an alternative test methodology is highly recommended.

CLINICAL TESTS SUPPORTING A DETERMINATION OF SUBSTANTIAL EQUIVALENCE

Correlation (Comparison) Studies

Three independent studies of the correlation of the IgM Dip-S-Ticks Leptospirosis Test with available serological methods were conducted. In these studies, the Microscopic Agglutination (MAT) method was considered as the reference method and primary basis for comparison.

The commercially available Indirect Hemagglutination (IHA) Test, although intended for the detection of both IgM and IgG antibodies, was also employed.

Paired acute and convalescent specimens, as well as single specimens, from patients having clinical symptoms consistent with leptospirosis were analyzed in all studies. Test comparisons were based on the expression of the number of true positive (TP), true negative (TN), false positive (FP and false negative (FN) scores on individual specimens, assuming that the comparison methods (MAT and IHA) represent the TP result. The relative sensitivity, relative specificity and 95% confidence intervals of the sensitivity and specificity of the IgM Dip-S-Ticks test was determined for each study. Studies in which the MAT test was performed were identified as disease-confirmed or presumptive, based on the Centers for Disease Control and Prevention (CDC) case definition for leptospirosis (5). These criteria identify disease-confirmed patients as having a fourfold or greater increase in antibody titer between acute and convalescent-phase specimens obtained equal to or greater than 2 weeks apart. Presumptive patients lack a fourfold increase in antibody titer but show a titer of equal to or greater than 200 in one more specimens.

In all studies, the comparison analysis of methods was based on the number of days post-onset that individual specimens were obtained from symptomatic patients. Table 3 summarizes specimens obtained less than 10 days, 10-30 days, greater than 30 days post-onset and all data. The number of true positive (TP), true negative (TN), false positive (FP) false negative (FN), relative sensitivity, relative specificity and 95% confidence intervals of the sensitivity and specificity are included for each group.

A domestic prospective study (Study No. 1) was conducted of single and paired specimens from 286 symptomatic patients at the Hawaii Department of Health, Honolulu, Hawaii. The MAT, IHA and IgM Dip-S-Ticks tests were employed in the analysis of all specimens. In this study, 30 patients were confirmed by the MAT method, with a 4-fold increase in antibody titer.

TABLE 3
COMPARISON OF METHODS
PROSPECTIVE STUDY IN HAWAII

METHODS COMPARED	DAYS POST ONSET	TP	TN	FP	FN	TOTAL	RELATIVE SENS. %	CONFIDENCE INTERVAL 95%	RELATIVE SPEC. %	CONFIDENCE INTERVAL 95%
IHA and	All Data*	29	467	39	2	537	93.5	78.6—99.2	92.3	89.6—94.5
IgM Dip-S-Ticks	<10 Days	3	182	17	0	202	100.0	29.2—100.0	91.5	86.7—94.9
	10-30 Days	20	129	11	2	162	90.0	70.8—98.9	92.1	86.4—96.0
	>30 Days	6	156	11	0	173	100.0	54.1—100.0	93.4	88.5—96.7
MAT and	All Data*	53	442	21	29	545	64.6	53.3—74.9	95.5	93.1—97.2
IgM Dip-S-Ticks	<10 Days	11	171	10	10	202	52.4	29.8—74.3	94.5	90.1—97.3
	10-30 Days	26	120	9	9	164	74.3	56.7—87.5	93.0	87.2—96.8
	>30 Days	16	148	3	9	176	64.0	42.5—82.0	98.0	94.3—99.6
IHA and MAT	All Data*	30	458	1	49	538	38.0	27.3—49.6	99.8	98.8—100.0
	<10 Days	3	179	0	20	202	13.0	10.6—15.4	100.0	98.0—100.0
	10-30 Days	20	128	1	11	160	64.5	45.4—80.8	99.2	95.8—100.0
	>30 Days	6	150	0	18	174	25.0	10.7—50.2	100.0	97.6—100.0

A retrospective study (Study No. 2) was conducted in Barbados, consisting of paired specimens from 51 symptomatic leptospirosis patients and paired specimens from 52 symptomatic patients diagnosed with febrile diseases other than leptospirosis.

TABLE 4
COMPARISON OF METHODS
RETROSPECTIVE STUDY IN BARBADOS

METHODS COMPARED	DAYS POST ONSET	TP	TN	FP	FN	TOTAL	RELATIVE SENS. %	CONFIDENCE INTERVAL 95%	RELATIVE SPEC. %	CONFIDENCE INTERVAL 95%
IHA and	All Data*	70	105	18	4	197	94.6	86.7—98.5	85.4	79.1—91.6
IgM Dip-S-Ticks	<10 Days	34	61	16	1	112	97.1	85.1—99.9	80.3	69.5—88.5
	10-30 Days	24	26	2	2	54	92.3	74.9—99.1	92.9	76.5—99.1
MAT and	All Data*	72	109	17	7	205	91.1	82.6—96.4	86.5	80.5—92.5
IgM Dip-S-Ticks	<10 Days	37	62	14	3	116	92.5	79.6—98.4	81.6	71.0—89.5
	10-30 Days	25	28	2	3	58	89.3	71.8—97.7	93.3	77.9—99.2
IHA and MAT	All Data*	62	110	10	15	197	80.5	69.9—88.7	91.7	85.2—95.9
	<10 Days	27	66	7	12	112	69.2	52.4—83.0	90.4	81.2—96.1
	10-30 Days	24	25	2	3	54	88.9	70.8—97.7	92.6	75.7—99.1

A retrospective study (Study No. 3) was conducted at the Institute for Hygiene and Tropical Medicine, Lisbon, Portugal. This study consisted of 69 paired specimens from symptomatic patients comparing the performance of the MAT and IgM Dip-S-Ticks tests.

TABLE 5
COMPARISON OF METHODS
RETROSPECTIVE STUDY IN PORTUGAL

METHODS COMPARED	DAYS POST ONSET	TP	TN	FP	FN	TOTAL	RELATIVE SENS. %	CONFIDENCE INTERVAL 95%	RELATIVE SPEC. %	CONFIDENCE INTERVAL 95%
MAT and	All Data*	65	63	7	2	137	97.0	89.6—99.6	90.0	80.5—95.9
IgM Dip-S-Ticks	<10 Days	11	24	3	2	40	84.6	54.5—98.1	88.9	70.8—97.7
	10-30 Days	29	19	1	0	49	100.0	88.1—100.0	95.0	75.1—99.9
	>30 Days	15	15	2	1	33	93.8	69.8—99.8	88.2	63.6—98.5

Note: Please be advised that "relative" refers to the comparison of this assay's results to that of a similar assay. There was not an attempt to correlate the assay's results with disease presence or absence. No judgment can be made on the comparison assay's accuracy to predict disease.

The results of the studies indicate a good relative sensitivity and specificity for the IgM Dip-S-Ticks Leptospirosis test when compared to the IHA and MAT tests. However, Study No. 1 showed a much better relative sensitivity for the comparison of IHA and IgM Dip-S-Ticks tests than for the comparison of the MAT and IgM Dip-S-Ticks and the MAT and IHA tests. A good relative specificity was also reported in these studies.

Culture Isolation Studies:

Leptospira organisms were cultured from the blood or urine of 12 patients in Hawaii and 24 patients in Barbados to confirm leptospirosis infections and to identify the infecting serovars. In Hawaii, 9 culture positive patients (75%) were IgM Dip-S-Ticks positive and 10 patients (80%) were MAT positive. In Barbados, all of 24 culture positive patients (100%) were IgM Dip-S-Ticks positive and 23 patients (95.8%) were MAT positive. Both tests demonstrated broad reactivity to infecting serovars as well as additional serovars identified by the MAT test but not isolated. The most common serovar isolated in the Hawaii study was australis and in the Barbados study the most common serovar isolated was bim. However, serovar expression may vary significantly over time and within a given region.

BIBLIOGRAPHY

1. Levett P.N., Whittington C.U., Evaluation of the indirect hemagglutination assay for the diagnosis of acute leptospirosis, J. Clin. Microbiol. 36:11-14, 1998.
2. Ribeiro M.A., Assis C.S.N., Romero E.C., Serodiagnosis of human leptospirosis employing immunodominant antigen, Serodiagn. Immunother. Infect. Disease, 6:140-144, 1994.
3. Turner L.H., Leptospirosis II, Trans. Royal. Soc. Trop. Med. & Hygiene, 62:880-889, 1968.
4. Pappas M.G., Ballou R., Gray M.R. et al, Rapid serodiagnosis of leptospirosis using the IgM-specific dot-ELISA: Comparison with the microscopic agglutination test, Am. J. Trop. Med. Hyg. 34(2) 346-354, 1985.
5. Case definitions for infectious conditions under Public Health Surveillance, MMWR 1997; 46(No.RR-10):49.
6. Sulzer C.R., Glosser J.W., Rogers F. et al, Evaluation of an indirect hemagglutination test for the diagnosis of leptospirosis, J. Clin. Microbiol. 2:218- 221, 1975.
7. NCCLS Guideline: Assessment of the clinical accuracy of laboratory tests using Receiver Operating Characteristics (ROC) plots; Approved Guideline GP10-A, Vol 15, No. 19, December 1995.
8. Ashford D., Epidemiology, Symposium titled Leptopsirosis, a disease that is re-emerging on a worldwide basis, 49th Annual Meeting of American Society of Tropical Medicine and Hygiene, Houston, Texas, October 29-November 2, 2000.

DEPARTMENT OF HEALTH & HUMAN SERVICES

Food and Drug Administration
2098 Gaither Road
Rockville MD 20850

NOV 22 2000

PanBio InDx, Inc.
c/o David C. Bishop, Ph.D.
222 Stonebrook Drive
Hendersonville, North Carolina 28791-1555

Re: K002024
Trade Name: PanBio InDx® Dip-S-Ticks® Leptospirosis Test for the Detection of IgM Antibodies to Leptospira biflexa
Regulatory Class: II
Product Code: GRY
Dated: September 21, 2000
Received: September 22, 2000

Dear Dr. Bishop:

We have reviewed your Section 510(k) notification of intent to market the device referenced above and we have determined the device is substantially equivalent (for the indications for use stated in the enclosure) to legally marketed predicate devices marketed in interstate commerce prior to May 28, 1976, the enactment date of the Medical Device Amendments, or to devices that have been reclassified in accordance with the provisions of the Federal Food, Drug, and Cosmetic Act (Act). You may, therefore, market the device, subject to the general controls provisions of the Act. The general controls provisions of the Act include requirements for annual registration, listing of devices, good manufacturing practice, labeling, and prohibitions against misbranding and adulteration.

If your device is classified (see above) into either class II (Special Controls) or class III (Premarket Approval), it may be subject to such additional controls. Existing major regulations affecting your device can be found in the Code of Federal Regulations, Title 21, Parts 800 to 895. A substantially equivalent determination assumes compliance with the Current Good Manufacturing Practice requirements, as set forth in the Quality System Regulation (QS) for Medical Devices: General regulation (21 CFR Part 820) and that, through periodic QS inspections, the Food and Drug Administration (FDA) will verify such assumptions. Failure to comply with the GMP regulation may result in regulatory action. In addition, FDA may publish further announcements concerning your device in the Federal Register. Please note: this response to your premarket notification submission does not affect any obligation you might have under sections 531 through 542 of the Act for devices under the Electronic Product Radiation Control provisions, or other Federal laws or regulations.

This letter will allow you to begin marketing your device as described in your 510(k) premarket notification. The FDA finding of substantial equivalence of your device to a legally marketed predicate device results in a classification for your device and thus, permits your device to proceed to the market.

If you desire specific advice for your device on our labeling regulation (21 CFR Part 801 and additionally 809.10 for in vitro diagnostic devices), please contact the Office of Compliance at (301) 594-4588. Additionally, for questions on the promotion and advertising of your device, please contact the Office of Compliance at (301) 594-4639. Also, please note the regulation entitled, "Misbranding by reference to premarket notification" (21CFR 807.97). Other general information on your responsibilities under the Act may be obtained from the Division of Small Manufacturers Assistance at its toll-free number (800) 638-2041 or (301) 443-6597 or at its internet address "http://www.fda.gov/cdrh/dsma/dsmamain.html".

Sincerely yours,

Steven Gutman

Steven I. Gutman, M.D., M.B.A.
Director
Division of Clinical Laboratory Devices
Office of Device Evaluation
Center for Devices and Radiological Health

Enclosure

510(k) NUMBER (IF KNOWN): K002024_____

DEVICE NAME: PanBio-InDx IgM Dip-S-Ticks Leptospirosis Test_____

INDICATIONS FOR USE:

The PanBio InDx® IgM Dip-S-Ticks® Leptospirosis test is a qualitative enzyme immunoassay that specifically detects IgM antibodies to *Leptospira biflexa* (serovar *patoc 1*). This test is presumptive for the laboratory diagnosis of leptospirosis. The IgM Dip-S-Ticks Test may be used to evaluate a single specimen or paired specimens to detect sero-conversion. The test is intended for use in serum, plasma, heparinized whole blood or finger stick capillary blood.

Woody Dubois

(Division Sign-Off)
Division of Clinical Laboratory Devices
510(k) Number K002024

(PLEASE DO NOT WRITE BELOW THIS LINE—CONTINUE ON ANOTHER PAGE IF NEEDED)

Concurrence of CDRH, Office of Device Evaluation (ODE)

Prescription Use (Per 21 CFR 801.109) X OR Over-The-Counter Use (Optional Format 1) _____